Amendments to the Claims

Claim 1 (Currently amended): A method for improving viral vector titer in ~~establishing~~ a vector packaging cell line comprising:

a) introducing a helper virus into a cell population to establish a packaging cell line, wherein said helper virus comprises retroviral genes encoding virion structural proteins for retrovirus assembly in combination with an internal ribosome entry site linked to a selection marker gene;

b) selecting for helper virus-containing cells from step (a) in which the helper virus DNA is not methylated by growing the cells in the presence of a selection drug ~~agent~~ thereby reducing the presence of methylated helper virus within said cell population to allow for increased helper virus gene expression, thereby improving viral titer;

c) transducing the packaging cell line with a viral vector comprising a packaging sequence and a gene of interest; and

d) collecting viral particles.

Claim 2 (Currently amended): The method of claim 1 wherein said step of selecting for helper virus-containing cells in which the helper virus DNA is not methylated comprises: positively selecting for cells containing functional helper virus ~~which is functional~~, wherein said selection is by antibiotic resistance.

Claim 3 (Cancelled)

Claim 4 (Currently amended): The method of claim ~~3~~ 2 wherein said antibiotic resistance selection is accomplished via expression of a selection marker gene operably linked to helper virus sequences in combination with ~~ligation of~~ an internal ribosome entry site ~~with a selection marker~~ so that drug selection ensures promoter function in said helper virus.

Claim 5 (Currently amended): The method of claim 1 wherein said viral titer achieves levels of 1.5×10^7 cfu/ml in the presence of antibiotic ~~resistant~~ resistance selection.

Claim 6 (Original): The method of claim 1 wherein said helper virus comprises at least one viral production gene operably linked to viral promoter sequence which is capable of being methylated.

Claim 7 (Original): The method of claim 6 wherein said viral promoter comprises a long terminal repeat.

Claim 8 (Previously presented): The method of claim 7 wherein said viral promoter sequence is the long terminal repeat of a retrovirus.

Claim 9 (Currently amended): The method of claim 1 wherein said growing cells in the presence of the selection ~~agent~~ drug positively selects for cells in which the 5′ long terminal repeat of the helper virus is not methylated.

Claim 10 (Currently amended): The method of claim 1 wherein said selection ~~selecting~~ for helper virus DNA not methylated is with an antibiotic product Phleomycin D1 commonly known as ~~accomplished by treating vector producer cells with Zeocin~~ ZEOCIN™.

Claim 11 (Withdrawn): The method of claim 9 wherein said inhibiting of methylation is accomplished by: a step selected from the group consisting of:

insertion of a demethylation fragment of murine thy-1 in front of the 5' long terminal repeat.

Claim 12 (Withdrawn): The method of claim 9 wherein said inhibiting of methylation is accomplished by: a step selected from the group consisting of:

immune response selection.

Claim 13 (Withdrawn): The method of claim 9 wherein said inhibiting of methylation is accomplished by: a step selected from the group consisting of:

design of synthetic viral promoters to omit methylation sites.

Claim 14 (Cancelled)

Claim 15 (Withdrawn): The method of claim 9 wherein said inhibiting of methylation is accomplished by: a step selected from the group consisting of:

antisense inhibition of the human methylate gene.

Claim 16 (Withdrawn): A helper virus nucleotide sequence comprising:

a packaging deficient nucleotide sequence which encodes one or more structural viral components necessary for assembling a viral capsid, a viral promoter sequence capable of becoming methylated and a marker selection gene placed so that active helper virus may be positively selected.

Claim 17 (Withdrawn): The nucleotide sequence of claim 16 wherein said viral promoter sequence comprises:

a long terminal repeat promoter sequence which has been modified to inhibit methylation.

Claim 18 (Withdrawn): The helper virus nucleotide sequence of claim 16 wherein said sequence includes the methylation fragment of murine thy-1 in front of the 5' long terminal repeat site.

Claim 19 (Withdrawn): The helper virus nucleotide sequence of claim 16 wherein said sequence includes an internal ribosome entry site with a selection marker downstream of a viral component encoding sequence so that selection ensures promoter function.

Claim 20 (Withdrawn): The helper virus nucleotide sequence of claim 16 wherein said internal ribosome entry site is a picornavirus internal ribosome entry site.

Claim 21 (Withdrawn): The helper virus nucleotide sequence of claim 16 wherein said marker selection gene is an antibiotic resistance marker.

Claim 22 (Withdrawn): The helper virus nucleotide sequence of claim 16 wherein said helper virus is pAM3-IRES-Zeo.

Claim 23 (Withdrawn): A vector packaging cell, said cell comprising a helper virus nucleotide sequence according to claim 16.

Claim 24 (Withdrawn): A vector producer cell comprising a helper virus nucleotide sequence according to claim 16 and a viral vector, said producer cell capable of assembling virions particles.

Claim 25 (Withdrawn): An infectious viral particle produced by the method of claim 1.

Claim 26 (Currently amended): A method for increasing the presence of viral titer produced by a vector packaging cell upon transfection with a viral vector comprising:
decreasing the amount of inactive helper virus present in said vector packaging cell by removing methlyated helper virus, thereby improving helper gene expression and virion production within a selected population of vector packaging cells ~~providing the selection of non-methylated helper virus-containing cells~~.

Claim 27 (Currently amended): ~~A~~ The method of claim 26 wherein said step of selecting against DNA methylation of ~~for non-methylated~~ helper virus comprises selecting for cells having a functional helper virus by exposing said cells to an antibiotic so that cells with methylated helper virus are killed.

Claim 28 (Currently amended): The method of claim 26 wherein said step of d

inactive helper virus comprises the steps of:

removing from a population of vector packaging cells, helper virus with 5' long terminal repeat methylation by introducing an antibiotic to said cells.

Claim 29 (Currently amended): The method of claim 28 further comprising the step of:

removing cells with inactivated virus by killing said cells with antibiotic ~~positive~~ selection.

Claim 30 (Cancelled)

Claim 31 (Currently amended): The method of claim ~~30~~ 29 wherein said removal is accomplished by a helper virus with a picarnovirus internal ribosomal entry site sequence followed by an antibiotic resistance gene ~~marker~~ at the 3' end of the env sequence of said helper virus.

Claim 32 (Currently amended): The method of claim 31 wherein said antibiotic resistance selection marker gene confers resistance to Phleomycin D1, a product commonly known as ~~is~~ ~~Zeocin~~ ZEOCIN™.

Claim 33 (Currently amended): The method of claim 27 further comprising inhibiting DNA methylation of a helper virus long terminal repeat by ~~a step selected from the group consisting of:~~ treating vector producer cells with 5-Aza Cytidine (5-AZA-C).

Claim 34 (Withdrawn): The method of claim 27 wherein said inhibiting of methylation is accomplished by a step selected from the group consisting of:

insertion of a demethylation fragment of murine thy-1 in front of the 5' long terminal repeat.

Claim 35 (Currently amended): The method of claim 27 wherein said selecting for ~~non-methylated~~ functional helper virus is accomplished by ~~a step selected from the group consisting of:~~ the operably linking a viral component to a selection gene by ligation of an internal ribosome entry site with a selection ~~marker~~ gene that confers resistance to a drug so that drug selection would ensure promoter function.